

A Comparative Study of the Morphology and Histochemistry of the Reptilian Adrenal Gland

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(Plates I-V; Text-figure 1)

INTRODUCTION

THERE is considerable evidence indicating the importance of the mammalian adrenal cortex in regulating certain aspects of protein, carbohydrate and electrolyte metabolism (Sayers, 1950). It would seem that any general concepts of adrenal histophysiology would be enhanced by comparative studies in all vertebrate classes. The function of the equivalent cortical tissue in the poikilothermous reptiles presents a challenging problem that has received little attention. Moreover, the morphology of the reptilian adrenal gland has not been adequately described for many common North American species. A basic knowledge of normal morphology should be a prerequisite for accurate interpretation of experimental modifications.

The purpose of this study is to describe the normal anatomy, histology, cytology and histochemistry of the adrenal glands of certain common North American representatives of the various orders of reptiles in order to determine the nature of the similarities and differences manifested by the adrenals. By this approach it was hoped that types especially suitable for future experimental studies would be discovered.

Knowledge of adrenal morphology in the Crocodylia is based primarily upon a brief anatomical and histological description of the adrenals of the alligator, *Alligator mississippiensis*, by Reese (1931). Lawton (1937), also, has described the gross anatomy and histology of the the alligator adrenal with special reference to the adrenal-autonomic complex and vascular supply. The histogenesis of the alligator adrenal has been described by Forbes (1940).

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There is a paucity of information regarding the chelonian adrenal. Present knowledge is based principally on the early description of the adrenal in the turtle, *Emys orbicularis*, by Vincent (1898) and the description of the development of the adrenals in the loggerhead turtle, *Caretta caretta*, by Kuntz (1912).

The adrenals of the Serpentes have received little attention. Present knowledge is based mainly on the early descriptions of colubrid snakes by Vincent (1898), Minervini (1904) and Radu (1934); recent observations by Valle & Souza (1942) on the histology of the adrenals in the snake, *Dryophylax* sp.; and brief notes on the morphology of certain South American snakes by Uchoa Junqueira (1944).

The adrenal morphology of the Sauria has received some attention and histological descriptions are available for a few species of lizards. The first histological description of the reptilian adrenal is that of the European lizard, *Lacerta agilis*, by Braun (1882). Kraus (1921) gave a rather complete histological description of this species. Bimmer (1950) has described the seasonal variations in the adrenals of the closely related lizard, *Lacerta vivipara*. Vincent (1896, 1898) described the histology of the adrenals in *Chameleon vulgaris*, *Anguis fragillis*, *Lacerta agilis* and *Uromastix hardwickii*. More recently, Retzlaff (1949) described the histology of the adrenals in the alligator lizard, *Gerrhonotus multicarinatus*. Hartman & Brownell (1949) gave a brief description of the adrenals in the Gila monster, *Heloderma suspectum*, and the iguanid, *Anolis carolinensis*. Perhaps the most complete discussion of reptilian adrenal morphology is that by Miller (1952), who described the normal histology of the viviparous lizard, *Xantusia vigilis*, and its experimental modification by hypophysectomy, starvation and the ad-

ministration of ACTH, Cortisone and DOCA; however, no histochemical studies were made.

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MATERIALS AND METHODS

The reptiles used in this study were obtained from various sources. Young alligators, *Alligator mississippiensis*, from Schriever, Louisiana, were obtained through local animal dealers. Horned lizards, *Phrynosoma cornutum*, were obtained from the vicinity of Abilene, Texas. Skinks, *Eumeces fasciatus*, were obtained from the vicinity of Mena, Arkansas. Painted turtles, *Chrysemys picta*, were collected in the vicinity of Lakehurst, New Jersey, and were also obtained from collectors near Oshkosh, Wisconsin. Garter snakes, *Thamnophis sirtalis*, were collected in the vicinity of Glen Harbor, Long Island, New York, and supplemented by collections obtained from the vicinity of Oshkosh, Wisconsin.

A total of 304 animals was used in the study. These may be classified under the following orders, families and species, conforming with the recent checklist of North American reptiles (Schmidt, 1953):

Order Crocodylia

Family Crocodylidae

- 16 *Alligator mississippiensis* Daudin, Alligator
- 10 *Caiman crocodilus* Linnaeus, South American Caiman

Order Chelonia

Family Emydidae

- 36 *Chrysemys picta* Schneider, Eastern Painted Turtle
- 12 *Pseudemys scripta* Schoepff, Terrapin
- 4 *Terrapene carolina* Linnaeus, Box Turtle
- 4 *Clemmys guttata* Schneider, Spotted Turtle

Family Chelydridae

- 6 *Chelydra serpentina* Linnaeus, Snapping Turtle

Family Kinosternidae

- 4 *Kinosternon flavescens* Agassiz, Mud Turtle

- 2 *Sternotherus odoratus* Latreille, Musk Turtle

Family Trionychidae

- 4 *Trionyx ferox* Schneider, Soft-shelled Turtle

Order Sauria

Family Iguanidae

- 36 *Phrynosoma cornutum* Harlan, Horned Lizard
- 12 *Anolis carolinensis* Voigt, Green Anolis
- 12 *Sceloporus undulatus* Latreille, Fence Lizard

Family Scincidae

- 24 *Eumeces fasciatus* Linnaeus, Five-lined Skink
- 6 *Eumeces obsoletus* Baird & Girard, Great Plains Skink
- 12 *Lygosoma laterale* Say, Little Brown Skink

Family Gekkonidae

- 1 *Tarantola mauritanica* Say, European Gecko
- 1 *Gonatodes fuscus* Hallowell, Yellow-headed Gecko
- 1 *Sphaerodactylus cinereus* Wagler, Ashy Gecko
- 4 *Hemidactylus turcicus* Linnaeus, Turkish Gecko

Family Anguidae

- 12 *Gerrhonotus coeruleus* Wiegmann, Alligator Lizard
- 6 *Ophisaurus ventralis* Linnaeus, Eastern Glass Snake

Family Teiidae

- 8 *Cnemidophorus tessellatus* Say, Checkered Race Runner

Order Serpentes

Family Colubridae

- 48 *Thamnophis sirtalis* Linnaeus, Eastern Garter Snake
- 6 *Natrix sipedon* Linnaeus, Common Water Snake
- 6 *Lampropeltis dolia* Lacépède, Eastern Milk Snake
- 6 *Opheodrys vernalis* Harlan, Smooth Green Snake

Family Crotalidae

- 2 *Crotalus viridis* Rafinesque, Prairie Rattlesnake

Family Boidae

- 2 *Boa constrictor* Linnaeus, Boa Constrictor
- 4 *Charina bottae* Blainville, Rubber Boa

The animals, upon arrival in the laboratory, were kept in large vivaria, given ample water and supplied with food. Every attempt was made to utilize them as soon as possible after arrival, and only those specimens appearing to be in a healthy condition were used for histological study. The animals were anesthetized by intraperitoneal injections of sodium Nembutal in a 1:100 solution in 0.9% saline. The adrenals and immediately-surrounding tissues were rapidly removed and placed in fixative. Routine fixation fluids included 10% neutral formalin, Bouin's and Zenker-formal.

Tissues for general histological study were dehydrated and cleared by the dioxane method (Lillie, 1948). The tissues were then imbedded in 56-58° C. Tissuemat, serial sectioned at 5, 7 and 10 microns and the sections attached to slides with Mayer's albumen glycerol (Lillie, 1948). The following stains were used routinely: Mayer's acid hemalum and eosin, Masson's trichrome stain and Heidenhain's modification of Mallory's azocarmine-aniline blue technique (Lillie, 1948).

The distribution of connective tissue fibers was studied by a number of methods. Collagen was demonstrated by the Masson trichrome stain and the Van Gieson picric-acid fuchsin collagen method (Lillie, 1948). Weigert's iron hematoxylin was used as a nuclear stain with both methods. Elastic fibers were stained by Verhoeff's elastic tissue stain (McClung, 1952). To demonstrate reticulum, the Foot modification of Hortege's silver carbonate method was employed (McClung, 1952). Nerve fibers within the adrenal gland were stained by the Pearson-O'Neill (1946) silver-gelatine method following Bouin fixation.

For the study of mitochondria, tissues were fixed either in Regaud's fluid for 4 days and post-chromated for 7 days in 3% potassium dichromate (Lillie, 1948), or by Zenker-formal fixation followed by post-chromation for 48 hours at 37° C. in 3% potassium dichromate. The tissues were then dehydrated in graded alcohols, cleared in cedarwood oil and transferred through xylol to paraffin. Sections were cut at 3 microns. Mitochondrial stains used included Regaud's hematoxylin, Heidenhain's iron hematoxylin and aniline-acid fuchsin-methyl blue (Lillie, 1948).

Histochemical studies of cortical lipids were carried out on tissues fixed for 3 days in 10% formal-calcium (Baker, 1944). The tissues were then embedded in gelatin (Zwemer, 1933) and sectioned on the freezing microtome at 10 and 15 microns. The sections were attached to gelatinized slides as recommended by Baker (1946) and stored in Baker's formal-calcium-cadmium

preservative. Histochemical tests were applied to the sections within a two-week period after the time of fixation. Prolonged storage in formalin resulted in a diffuse background staining.

The sudanophilic lipids of the cortical cells were demonstrated by Sudan black B in propylene glycol without counterstaining (Pearse, 1952). Cholesterol and its esters were demonstrated by the Shultz test (Lillie, 1948). The distribution of carbonyl-containing lipids was investigated by means of the Schiff leucofuchsin reagent (Lillie, 1948).

OBSERVATIONS

A. *Crocodilia*

(a) *Alligator mississippiensis*

Anatomy

The adrenals of the young alligator, *Alligator mississippiensis*, are elongate bodies located in a retroperitoneal position on the midventral surface of the kidneys and extending slightly anterior to the latter with the right adrenal extending slightly more cephalad than the left. They are in close association with the ventrally situated gonads and the laterally placed gonaducts. The adrenal gland is a discrete structure, separated from the kidney and reproductive organs by a distinct connective tissue capsule. The medial surface of the adrenals borders on the post caval vein (Fig. 1).

In shape, the adrenals appear ellipsoid in longitudinal view and ovoid-to-round in cross-section. The surface of the gland is smooth. In animals ranging in body length from 60 to 75 cm., the glands vary in size from 10 to 12 mm. in length and from 2 to 3 mm. in width. They can be distinguished from the gonads and kidney by their pinkish-white to yellowish-orange color.

The vascular supply consists of both arterial and venous blood. Several adrenal arteries branch off each side of the dorsal aorta and ramify over the dorsal surface of the gland in the connective tissue capsule. Small arterioles branch off and penetrate the parenchyma of the gland, finally connecting with venous sinuses. Arising from the dorsal body wall lateral to the midline, a medium-sized afferent vein enters the anterior dorsal surface of the adrenal and finally branches into a capsular network. The right adrenal is in intimate contact with the vena cava with a large central venous sinus of the gland opening directly into the vena cava through a series of openings or ostia in the wall of the vena cava. The left adrenal gathers blood into a large ventro-mesial capsular vein which empties into the vena cava near the center of the adrenal gland (Fig. 1).

The adrenal is innervated from the paired lateral sympathetic ganglia in the region of the

kidney. Sympathetic nerve fibers from at least four sympathetic ganglia supply each adrenal, thus producing a more or less segmented innervation. Large nerves consisting principally of unmyelinated fibers ramify through the pericapsular region (Fig. 2). In frozen sections stained with Sudan black B, thin rings of sudanophilic material are often noticed in the nerve fiber bundles. The nerves appear to be encapsulated in a sheath of collagenous fibers. Sympathetic ganglion cell clusters are located on the dorsal side of the gland in the connective tissue capsule. Following silver impregnation, a network of argyrophilic fibers can be distinguished. Coarse nerve fibers tend to follow small blood vessels toward the center of the gland and then branch out to form networks over the surface of cortical cords and medullary cell masses. From these networks fine thread-like fibers branch off and penetrate between the cortical and medullary cells, terminating as very fine threads.

Histology

The stroma of the adrenal consists of a distinct connective tissue capsule composed of collagenous and reticular fibers. The collagenous fibers are continuous with the surrounding connective tissue while the reticular fibers are more or less limited to the inner zone of the capsule and are continuous with the parenchymal reticulum. An inner parenchymal meshwork of reticular fibers supports the cortical cords and medullary cells (Fig. 3). While collagenous fibers are the predominant type found in the capsule, occasional short elastic fibers are noticed in the walls of capsular veins. No muscle tissue, other than that occurring in the media of small arteries, is present. Infrequent bundles of collagenous fibers form trabeculae which penetrate into the center of the gland carrying arterioles and nerve fibers. Fine collagenous fibers course between the parenchymal cell cords as well as in the walls of the vascular sinuses. Heavy reticular fibers in the inner layers of the capsule extend inward and provide an extensive network to support the cortical cells and medullary masses. Coarse fibers, stretching lengthwise between the cortical cords, give off a network of fine reticular fibers at right angles which enclose the cortical cords and medullary cell masses in a reticular framework (Fig. 3).

The alligator adrenal is a composite endocrine gland composed of cortical and medullary cells. The medullary tissue consists of a peripheral layer that encircles the cortical tissue together with small clumps and strands of medullary cells scattered throughout the interspaces between the cords of cortical cells and the vascular sinuses (Fig. 2). Intertwining and an-

astomosing solid cords measuring from 25 to 35 microns in diameter constitute the cortical tissue. Occasional cortical cords extend through the peripheral medullary tissue and touch upon the capsular connective tissue. Small nodules of undifferentiated cortical cells are often found scattered in the capsule. An intricate network of capillaries and venous sinuses pervades the cortical and medullary tissue.

The zonation typical of the mammalian adrenal is lacking in the alligator adrenal. The peripheral cords tend to be made up of elongated cells in a more or less radial arrangement while the central region of the gland consists of cords made up of irregularly dispersed polyhedral cells having centrally placed nuclei. These central cords often appear to be oriented toward the side of the gland bordering on the vena cava (Fig. 1). The venous sinuses are lined with an endothelium having flattened granular nuclei.

Cytology

The medullary tissue is composed of two cell types with different tinctorial and morphological characteristics. These differences are best observed after Zenker-formal fixation and staining with Masson's or with Heidenhain's azan stain. A peripheral zone of yellow-staining cells can be readily distinguished from the more centrally situated reddish-brown cells (Fig. 2). The yellow-stained cells will be referred to as the "light" cells and the reddish-brown cells as "dark" cells. The "light" cells are 15 to 18 microns in diameter with a centrally located nucleus measuring 5 to 7 microns and are spherical to polyhedral in shape. These "light" cells have a finely granular cytoplasm and occur singly, in small clumps and strands, and in rather large masses in the peripheral regions of the adrenal (Fig. 2). The majority of them are restricted to the capsular region; however, occasional strands extend inward among the cortical cords, especially in the connective tissue trabeculae penetrating the gland. The "light" medullary cells are often found in association with sympathetic ganglion cells in the capsule or in the adjacent connective tissue.

The "dark" cells occur singly or in small clumps or strands in the spaces and sinuses which pervade the cortical tissue (Figs. 2 & 5). This close association of "dark" medullary cells with the cortical tissue is strikingly evident in all tissues examined. These cells are, as a rule, smaller in size, measuring from 10 to 12 microns, and are generally polyhedral in shape. The granules in the cytoplasm are coarser and appear reddish-brown after staining with Masson's. The nucleus measures from 4 to 5 microns in diameter and is spherical, with 1 or 2 nucleoli.

In sections, the cords of cortical cells appear as spherical-to-elongate anastomosing masses of cells usually consisting of 1 to 4 tiers of cells (Fig. 2). The shape of the cells in the cord varies from irregular to greatly elongated spindle-shaped or wedge-shaped. The latter configurations are probably due to compression forces, since they are found more often in the tips of peripheral cords abutting against the capsule. The nuclei are centrally located, measuring 5 to 6 microns in diameter, and usually possess only one large fuchsinophilic nucleolus. Following Zenker-formal fixation and Masson's or Heidenhain's azan stain, the cytoplasm has a slightly fuchsinophilic and finely reticulated appearance.

Mitochondria are best demonstrated after post-chromation of Zenker-formal fixed material and staining with aniline-acid fuchsin and methyl blue. The small, granular, red-staining mitochondria are scattered throughout the cytoplasm (Fig. 5). The amount and distribution of mitochondria varies with the lipid concentration in the cortical cells. Lipid-poor cells exhibit a strong fuchsinophilia with the mitochondria concentrated in the perinuclear zone. Lipid-rich cells have granular mitochondria dispersed in the cytoplasmic matrix between the lipid vacuoles. One to several relatively large fuchsinophilic spheres are often seen in the cytoplasm in the vicinity of the nucleus.

In Baker's formal-fixed frozen sections stained with Sudan black B, the medullary cells and surrounding tissue remain colorless in contrast to the intense blue-black staining of the cortical cells (Fig. 4). The sudanophilic lipids are evenly distributed throughout the cortical tissue. Under high magnification the sudanophilic droplets appear to be more or less evenly distributed throughout the cytoplasm. There are differences in the amount of sudanophilic substances present: small undifferentiated cells possess little sudanophilic material, whereas other cells (particularly in peripheral cords) are filled with sudanophilic droplets.

Frozen sections subjected to the Shultz cholesterol test develop an intense blue-green color in the cortical tissue only. Medullary tissue, connective tissue and kidney tissue give a negative reaction to this procedure. The Shultz-positive droplets have a uniform distribution throughout the cytoplasm of the cortical cells, similar to the sudanophilic droplets. An intense and selective staining of frozen sections is observed upon their treatment with Schiff leucofuchsin. A purplish-red color develops throughout the cortical tissue. Except for the interesting observation that the elastic fibers of blood vessels stain a purple color, all other tissues in the sec-

tions are Schiff-negative. The distribution parallels that of the sudanophilic lipids.

(b) *Caiman crocodilus*

The animals used in this study were very young specimens, still having an internal remnant of the yolk-sac and a visible umbilical scar on the abdomen. The adrenal of the immature caiman is similar in anatomical and histological features to the adrenal of the alligator. The only significant difference noted was in connection with the cortical tissue. The cortical cells are not as well differentiated in the center of the gland as are the peripheral cells. The centrally located cells are small and polyhedral and irregularly arranged in cords, while those at the periphery are of a tall columnar type.

B. *Chelonia*

1. Family Emydidae

(a) *Chrysemys picta*

Anatomy

The adrenal glands of the painted turtle, *Chrysemys picta*, are paired bodies, yellowish in color, extending along the ventro-mesial surface of the kidneys in a retroperitoneal position (Fig. 6). They have an irregular outline, often appearing to consist of partially-fused spherical or irregularly-shaped masses imbedded around the numerous efferent renal veins. The gland is not completely encapsulated, but there is a close association and often an intermingling of renal tissue and adrenal tissue on the dorsal side of the adrenal. The ventral surface of the gland is covered by the pleuroperitoneum which, at the ventro-lateral edge, gives rise to the mesentery supporting the gonad.

There is an interesting difference in the anatomical relationships of the kidney, adrenal and gonad in the turtle, when compared with those of the alligator. The gonad of the young alligator is attached to the ventral surface of the adrenal, whereas the gonad in the turtle is separate and suspended in a mesenteric fold of the pleuroperitoneum (Figs. 1 & 6).

The arterial blood supply consists of several small adrenal arteries branching off from the dorsal aorta, as well as small branches arising from the genital and renal arteries that border or penetrate through the adrenal. Because of the unique location of the turtle adrenal with respect to the blood vessels of the kidney, it is difficult in histological sections to determine the identity of the veins and venous sinuses penetrating through the parenchyma of the gland. Doubtless, most of the sinuses represent efferent renal sinuses; however, some may be either sinuses of the renal portal veins or sinuses of afferent adrenal veins, all of which constitute a complex venous plexus in the region of the adrenal.

Numerous sympathetic nerve ganglia are found in the turtle adrenal, occurring most often in the connective tissue between the adrenal and the kidney or in the trabeculae which penetrate into the adrenal (Fig. 8). Following the silver-gelatine technique, a dense network of nerve fibers can be distinguished surrounding the medullary and cortical masses. A heavy network of fibers encircles and penetrates among the medullary cells, while in contrast only occasional fine, faintly-stained fibers penetrate the cortical tissue.

Histology

The stroma of the turtle adrenal is similar to that described for the alligator, except that in the turtle the gland is not completely encapsulated. A distinct capsule composed of collagenous and reticular fibers covers the ventral surface. Numerous trabeculae of collagenous and reticular fibers extend into the parenchyma, often appearing to separate the gland into separate masses of tissue. The columns and clumps of cortical and medullary tissue are outlined by a dense network of reticular fibers which serve as a framework to maintain the structural unity of the gland. Elastic fibers are limited to the adventitia of arteries and veins occurring in the capsule and trabeculae.

The adrenal of the painted turtle consists of intermixed cortical and medullary tissue (Fig. 7). The medullary tissue is not concentrated in any particular region of the gland, but is irregularly distributed among the cortical tissue as clusters of medullary cells. These clusters are of irregular size and shape, varying from small groups containing a few cells to large clumps and strands. It often appears that these clusters and strands lie in the vascular sinuses (Fig. 7).

The cortical tissue is composed of anastomosing and intertwining columns varying in width from approximately 40 to 70 microns. The columns are made up of several tiers or rows of cells which usually have an irregular arrangement (Figs. 7 & 8). Sometimes the cells in peripheral cords in longitudinal section have the appearance of being compressed into a single row of greatly elongated and flattened cells (Figs. 7 & 10).

Cytology

The medullary tissue consists of two cell-types which can be distinguished principally by their staining properties (Fig. 7). The peripheral medullary cells, after Zenker-formal fixation and Masson's stain, have a light brownish-yellow color in contrast to the darker yellowish-brown cells found in the more central region of the gland. The "light" cells occur singly or in small clumps or strands in the peripheral cap-

sular region, and sometimes extend for a short distance in toward the center of the gland in the intercortical spaces, especially in trabeculae that penetrate the gland. The "dark" cells occur singly or in small clumps or strands either imbedded in the side of cortical cords or in the intercortical spaces and sinuses.

A striking histological feature of the "dark" cells is the fact that they are usually in close contact with cortical tissue. On the other hand, the "light" cells are more often separate from the cortical tissue, either in the peripheral capsule or in the surrounding connective tissue. They are also found in association with sympathetic ganglia (Fig. 8), in the adventitia of the dorsal aorta and in the parenchymal connective tissue of the kidney. Both cell types are polyhedral-to-spherical in shape, varying from 9 to 12 microns in diameter. The nucleus varies in size from 5 to 7 microns.

The cortical cells are variable in size and shape. The most common morphological type is an irregularly-shaped polyhedral cell (Figs. 7 & 10). Sometimes, due to compression forces on the column, the cells assume a tall, columnar-to-spindle-shaped form (Fig. 10). The centrally located nuclei are 5 to 6 microns in diameter, are slightly reticulated and usually have a single large hyaline nucleolus. Fuchsinophilic spheres are commonly found in a juxtannuclear position.

The mitochondria appear as small granules scattered throughout the cytoplasm, with the number of mitochondria depending upon the amount of lipid in the cell (Fig. 10). Highly vacuolated cells have few mitochondria, whereas cells having little lipid are mitochondria-rich. Only an occasional mitotic figure was noted in the cortical cells. The lack of any concentration of undifferentiated cells in the columns and a random dispersion of mitotic figures suggests that cell replacement must take place within the cords in a random fashion.

The cortical cell lipids are sudanophilic, Shultz-positive and Schiff-positive, with a uniform and parallel distribution throughout the cortical tissue. Under high magnification, the sudanophilic droplets appear to be evenly distributed throughout the cytoplasm and no zonation of the gland is seen (Fig. 9).

(b) *Pseudemys scripta*, *Clemmys guttata*, *Terrapene carolina*

The adrenals of the other members of the family Emydidae included in this study, namely, *Pseudemys scripta*, *Clemmys guttata* and *Terrapene carolina*, resemble (both anatomically and histologically) the descriptions for *Chrysemys picta*. The only significant histological difference is found in the adrenals of the spotted

turtle, *Clemmys guttata*. In this species there is considerably more medullary tissue. In transverse sections, the intermingling clumps and strands of medullary and cortical cells appear to be approximately equal in size and number.

2. Family Chelydridae

The adrenals of the snapping turtle, *Chelydra serpentina*, show no significant morphological differences from the description of *Chrysemys picta*.

3. Family Kinosternidae

The morphology of the adrenals of the mud turtle, *Kinosternon flavescens*, and the musk turtle, *Sternotherus odoratus*, resembles that of *Chrysemys picta* in general features. The glands in transverse sections appear more elongated and closely associated with the kidneys. The medullary tissue is often located in the adjacent renal tissue as yellow clumps among the renal tubules. The cortical cells show a tendency toward a regular arrangement in cords measuring approximately 50 microns in diameter. The cells are tall, columnar in shape and usually radially arranged in the cords so that in longitudinal sections they appear as a double row of cells. The nuclei are usually located near the end of the cell distal to the vascular sinus.

4. Family Trionychidae

The adrenals of the immature soft-shelled turtle, *Trionyx ferox*, resemble the adrenals of immature *Chrysemys picta* both in anatomical and histological features. In young animals such as these, the cortical cells are grouped in narrow cords and are not well differentiated because they have little cytoplasm or lipid material.

C. Sauria

1. Family Iguanidae

(a) *Phrynosoma cornutum*

Anatomy

The adrenal glands of the horned lizard, *Phrynosoma cornutum*, are paired bodies located near the anterior ends of the gonads and enclosed in the mesorchia or mesovaria supporting the gonads. In the male, the adrenals are intimately associated with the sperm ducts (Fig. 11). The right adrenal is slightly cephalad to the left, extending along the vena cava to the right lobe of the liver.

The glands are elongate, oval-shaped bodies measuring approximately 2 to 3 millimeters in length and 1 millimeter in width. The combined weight of the freshly-dissected glands in adult animals is approximately 1 milligram.

The blood supply consists of both arterial and venous blood. Each adrenal receives a branch

from the dorsal aorta. These branches arise from the aorta at the origin of intervertebral arteries. In some specimens, the left adrenal receives two arteries from the aorta. A small afferent vein arises from the dorsal body wall cephalad and enters the anterior pole of the gland. The left adrenal is in contact with the cephalad end of the left efferent renal vein into which blood from the gland drains directly. The right adrenal is situated at the junction of the efferent renal veins and anteriorly along the vena cava. Blood from this gland drains directly into the vena cava.

Histology

The adrenal gland is enclosed in a thin connective tissue capsule composed of an outer layer of collagenous fibers which are continuous with the surrounding connective tissue, and an inner layer of reticular fibers which are continuous with the reticular framework supporting the parenchyma of the gland. Numerous trabeculae containing collagenous fibers and reticular fibers penetrate the parenchyma (Fig. 12).

Sympathetic ganglion cells are aggregated principally in the dorsal region of the capsule near the adrenal artery (Fig. 12). Occasional sympathetic cells are found in association with groups of medullary cells in other regions of the capsule. The adrenal gland of the horned lizard consists of cortical and medullary tissue arranged in a typical saurian pattern (Fig. 12). The medullary component comprises an incomplete dorsal capsule of primarily "light" type medullary cells with a few strands often extending inward from the periphery. In addition, small groups and individual cells of the "dark" type are interspersed in the parenchyma.

The cortical tissue consists of intertwining and anastomosing cords measuring 30 to 50 microns in diameter (Fig. 12). In transverse section, a typical cord appears as a radially arranged group of 15 to 20 tall columnar cells with very indistinct cell outlines. The nuclei are regularly arranged in a zone near the periphery of the cord. In longitudinal section, a cord often presents the characteristic appearance of a double row of tall columnar cells with nuclei arranged in a double row near the peripheral edge of the cord. The central area of the cord, following Masson's staining, has a pale fuchsinophilic vacuolated appearance.

The cephalic pole of the right adrenal is often fused with the caudal tip of the dorsal lobe of the liver, both tissues being in contact with and partially encapsulating the vena cava in this region. The liver parenchyma is not distinctly separated from the cortical tissue by a connective tissue capsule and as a result small groups

of cortical cells in many sections appear to be isolated in the liver parenchyma.

Cytology

The medullary tissue consists of two cell-types (Fig. 12). The strands and masses of medullary cells in the peripheral capsular region are of the "light" type, having few granules and appearing light-yellow after Zenker-formal fixation. The medullary cells in the central part of the parenchyma are of the "dark" type, staining a reddish-brown after the Masson or azan technique. Both cell types are similar in their morphology, consisting of polyhedral cells which are 9 to 12 microns in diameter. The centrally located nucleus varies from spherical to oval in shape and measures 4 to 5 microns in diameter and has a granular appearance with one or more nucleoli. Granular mitochondria are dispersed throughout the cytoplasm.

The anastomosing cords of cortical tissue are composed of polyhedral cells of varying height (Fig. 12). The most frequently observed type is a tall, wedge-shaped columnar cell approximately 20 microns in height and 7 microns in width at the base. The spherical-to-oval-shaped nucleus is 4 to 5 microns in diameter and contains one or more nucleoli. Generally, only a single large fuchsinophilic nucleolus is present.

The mitochondria in the cortical cells appear as fine granules, either concentrated in the perinuclear zone or dispersed in the delicate cytoplasmic network surrounding the lipid vacuoles. The lipid droplets are evenly distributed throughout the cytoplasm and give a strong, positive color reaction to the Sudan procedure (Fig. 13). There is a general and parallel distribution of sudanophilic lipids, carbonyl lipids and cholesterol esters.

(b) *Anolis carolinensis*

The adrenals of the green anolis, *Anolis carolinensis*, are similar in anatomical features to *Phrynosoma* (Fig. 14). The histological picture differs somewhat in that the cortical cords appear to be narrower (25 to 35 microns) and are made up of radially arranged low columnar cells. The nucleus is centrally located or may, in some cords, be located near the edge of the cell bordering on the blood supply. Fewer medullary cells are seen in the parenchyma of the gland with most of them being concentrated on the dorsal surface of the gland.

(c) *Sceloporus undulatus*

The adrenals of the fence lizard, *Sceloporus undulatus*, resemble, in general, those of *Anolis*. In this species the cortical cells are of a low columnar type with a tendency toward exhibiting a polarity by the display of nuclei at the peripheral edges of the cortical cells.

2. Family Scincidae

Anatomy

The anatomical relationships of the adrenals in the three species of skinks that were studied, namely, *Eumeces fasciatus*, *Eumeces obsoletus* and *Lygosoma laterale*, are in general similar to the saurian pattern described previously for the iguanid lizard, *Phrynosoma*. The glands are elongate bodies closely associated with the gonads and gonaducts. The right adrenal in this group of lizards does not make contact or fuse with the dorsal lobe of the liver; however, the mesentery of the right gonad is continuous with the mesentery supporting the dorsal lobe of the liver. A single afferent adrenal vein enters the caudal pole of each adrenal. A large sympathetic ganglion is located near the cephalic pole of each adrenal.

Histology

The gland is enclosed in a thin capsule continuous with the surrounding mesentery and connective tissue. The parenchyma of the gland is supported by a network of thick reticular fibers.

The disposition of the cortical and medullary tissue corresponds to the saurian pattern described for *Phrynosoma*. The medullary cells form a partial capsule around the dorsal surface of the cortical tissue. Some medullary tissue in the form of individual cells and small clumps is diffused throughout the parenchyma. The medullary cells are of two tinctorial types: the "light" cells which are confined chiefly to the periphery; and the deeper-lying cells which are usually of the "dark" type.

The arrangement of the cortical cells within the cords is a unique and significant feature in the Scincidae (Figs. 18 & 23). In transverse section, a cord consists of 10 to 12 tall, wedge-shaped columnar cells arranged in a radial pattern with the nuclei abutting against the peripheral edge of the cell. In longitudinal section, the cord appears to consist of two rows of tall columnar cells with parallel rows of nuclei at the edges of the cord. This marked polarity of the cortical cells is present in all three species of skinks examined. Occasionally, a lumen is observed in the center of a cord, giving the cord an appearance that resembles an exocrine gland. These central cavities are not a consistent feature and probably are artifacts resulting from shrinkage.

Cytology

The cortical cells display a marked polarity with the nucleus abutting against the edge of the cell facing the vascular supply. The region of the cell distal to the nucleus contains a faintly-

staining cytoplasmic network surrounding lipid vacuoles of varying size. Following aniline-acid fuchsin and methyl blue, the red-staining mitochondria are found widely scattered in the cytoplasmic network (Fig. 19). The typical cortical cell is a lipid-rich, mitochondria-poor type. Undifferentiated cortical cells are not commonly seen in the capsule, which would seem to suggest that cortical cell replacement occurs within the individual cords and not by migration.

The unique polarity of the cortical cells is clearly demonstrated by histochemical tests of the lipid droplets. In frozen sections where a cord happens to be cut lengthwise, the blue-black sudanophilic lipids are confined to a central zone of the cord and the nuclear zone at the periphery fails to stain (Fig. 20). A similar pattern of distribution of Shultz-positive and Schiff-positive droplets is found. No significant difference in the uniformity of staining is apparent. The medullary and surrounding connective tissue do not give a positive reaction to any of the tests applied.

An intense blue-green color reaction is obtained by the Shultz test for cholesterol and its esters. The distribution of the blue-green droplets is localized in the cytoplasm distal to the nucleus. Following application of the Schiff leucofuchsin technique, an intense red color develops exclusively in the cortical tissue. The intensity and specific localization of the color reaction in the cytoplasmic region of the cortical cells indicates that the cortical tissue, following Baker's fixation, contains a high level of carbonyl lipids.

3. Family Gekkonidae

A limited number of specimens from this family was available for histological study. The series consisted of a single adult European gecko, *Tarantola mauritanica*; one adult yellow-headed gecko, *Gonatodes fuscus*; one adult ash gecko, *Sphaerodactylus cinereus*; and four adult Turkish geckos, *Hemidactylus turcicus*. The anatomical relationships in these specimens resemble the general saurian pattern already described. The disposition of the cortical and medullary tissue is similar to that noted in other groups of lizards.

A special and significant feature of the gecko adrenal is the extreme polarity of the cortical cells (Figs. 21 & 22). The arrangement of the cells in the cords is similar to that described for the Scincidae. The cortical tissue, in transverse or longitudinal sections, appears as spherical-to-oblong anastomosing cords measuring approximately 50 microns in diameter. The cells in longitudinal sections of a cord appear as tall, wedge-shaped columnar cells with their nuclei

abutting on the peripheral edges of the cells facing a capillary or venous sinus. Because of the extremely small size of these animals and the consequent small size of the gland, a typical transverse section appears to consist of 3 or 4 anastomosing cords.

A single specimen of the Dibamidae, *Dibamus novae-guineae*, and two representatives of the Pygopodidae, *Lialis burtoni* and *Pygopus lepidopus*, were examined histologically and found to have a gekkonid arrangement of the cortical cells.

4. Family Teiidae

The anatomical features of the adrenals of the checkered race runner, *Cnemidophorus tessellatus*, conform to the saurian pattern already described for *Phrynosoma*. The microscopic anatomy, however, shows certain characteristics which are distinctive of this species (Fig. 17). The medullary tissue is more widely dispersed within the cortical tissue. In transverse sections, a dorsal aggregation is present, as well as numerous clumps scattered throughout the parenchyma of the gland. Both "light" and "dark" cells are present with the "light" cell type more or less restricted to the dorsal aggregation or to extra-adrenal locations.

The cortical cells do not exhibit a consistent regular arrangement, but generally appear to have a more or less random dispersion (Fig. 17). Occasional cords, 20 to 30 microns in diameter, present a radial and polarized arrangement of tall columnar cells with nuclei aligned on the periphery. Most of the cells are low columnar to polyhedral in shape, measuring approximately 10 microns in height and 7 microns in width. The nuclei are usually centrally located or are found toward the edge of the cell facing a vascular sinus.

5. Family Anguinae

(a) *Gerrhonotus coeruleus*

The morphology of the adrenals of the alligator lizard, *Gerrhonotus multicarinatus*, has been described by Retzlaff (1949). They exhibit the general saurian anatomical relationships observed for other lizards. The significant and specific characteristic feature of the anguimorphid type of adrenal is illustrated by *Gerrhonotus coeruleus*. The cortical cells are irregularly shaped and do not arrange themselves in definite cords or columns (Fig. 15). Only cortical cells bordering on a vascular sinus tend to arrange themselves in a regular fashion around the sinus with the nuclei near the side of the cell which faces the sinus. Most of the cells have centrally located nuclei. An unusual feature of the cortical tissue of *Gerrhonotus* is the presence in large individuals of a narrow peripheral zone of cor-

tical cells that stain less intensely than the rest of the cortical cells (Fig. 15). No histochemical tests were made on the few animals available to determine the nature of the lipids in this peripheral zone.

(b) *Ophisaurus ventralis*

The adrenals of the glass snake, *Ophisaurus ventralis*, exhibit the general saurian pattern. In this species, the body is greatly elongated like that of a snake, yet the adrenals and associated reproductive organs display only a slight asymmetry—the right gonad being slightly more cephalad than the left, but no more so than the usual disposition in the Sauria. The microscopic anatomy of this species is similar to the anguid lizard, *Gerrhonotus*, just described.

D. Serpentes

1. Family Colubridae

(a) *Thamnophis sirtalis*

Anatomy

The adrenals of the Eastern garter snake, *Thamnophis sirtalis*, are discrete bodies closely associated with the asymmetrically-located gonads (Fig. 24). They can be distinguished from the latter by their golden-yellow color and difference in texture. The left adrenal lies anterior to the left kidney near the cephalic end of the left efferent renal vein and extends anteriorly for a short distance alongside the gonad. With reference to the ventral scutes or gastrosteges, the left adrenal lies approximately between the 31st to 34th gastrostege in females and between the 35th to 37th gastrostege in males, counting anteriorly from the anal plate. The right adrenal lies alongside the vena cava just anterior to the junction of the efferent renal veins and is closely associated with the caudal end of the right gonad. The right adrenal lies approximately between the 45th and 48th gastrostege in both sexes. The glands are contained in the mesovaria or mesorchia supporting the gonads (Fig. 24). In the male, there is a close association with the vas deferens.

The adrenals are elongate bodies varying in length from 5 to 15 mm., depending on the body length of the snake. Usually the right adrenal is slightly longer than the left. Occasionally a gland appears to be constricted into a bilobed shape. In transverse section, the gland is elliptical-to-round with a diameter of 1 to 3 millimeters. Adrenal weights for a series of 13 garter snakes of mixed sex range from 0.03 to 0.08 per cent. of the body weight, with an average of 0.045 per cent.

The garter snake adrenal is an extremely vascular organ having both an arterial and a venous blood supply. The dorsal aorta gives off to each

adrenal a branch (the spermatic or ovarian artery) which enters the mid-dorsal region of the gland, bifurcates, sending a branch anteriorly through the dorsal pericapsular connective tissue, and continues into the testes or ovary (Fig. 24). The posterior branch continues caudally through the dorsal pericapsular layer of the adrenal and finally joins the gonaduct arteries. Numerous arterioles branch off from these longitudinal arteries to enter the parenchyma of the gland.

A separate venous supply can readily be seen on gross examination. Two (sometimes only one) afferent veins emerge from the dorsal body wall slightly lateral to the midline and enter the mid-dorsal region of the gland. Usually the second vessel emerges from the body wall just posterior to the adrenal and joins a longitudinal vein in the mesotuberium which connects with the anterior branch from the body wall. Branches from this venous supply connect with the venous sinuses and capillaries in the parenchyma of the gland.

The blood from the left adrenal gland is carried by a number of small, extremely short, efferent adrenal veins opening into the left efferent renal vein. The blood from the right adrenal drains into the postcaval vein either by means of numerous short efferent veins or is so intimately associated with the vein that blood from the parenchymal sinuses drains directly into the vena cava through small openings (Fig. 24).

A large aggregation of sympathetic ganglion cells is located in the dorsal capsular region, usually near the artery. Occasional sympathetic nerve cells are seen in the capsule of the gland; however, they were not found in the parenchyma of the gland. The medullary cells are innervated by an extensive network of nerve fibers, although nerve endings to the cortical cells cannot be definitely distinguished in the silver carbonate preparations.

Histology

The garter snake adrenal, fully encapsulated by the connective tissue, is almost completely enveloped in the surrounding mesentery (Figs. 24 & 25). The capsule consists chiefly of collagenous fibers with a few fine reticular fibers in the innermost layers. The parenchyma of the gland is supported by a thick network composed primarily of collagenous fibers. Fine reticular fibers are found in the walls of the vascular sinuses, where they form a fine network enveloping the cords of cortical tissue.

The medullary and cortical tissues are disposed in a pattern somewhat similar to that of the lizards (Fig. 25). The greater part of the medullary tissue is concentrated in the dorsal

pericapsular region in the vicinity of the spermatogenic or ovarian artery. In transverse section, a large mass of "light" yellow cells is seen aggregated around the dorsally located artery. Medullary cells of the "dark" type are found scattered in and among the cords of cortical cells singly or in small groups of three or four cells (Figs. 25 & 28).

The cortical tissue exhibits a unique and characteristic arrangement (Figs. 25 & 28). The cortical cells are arranged in anastomosing and intertwining cylindrical cords measuring 40 to 50 microns in diameter. In transverse section, a cord appears spherical and to consist of 12 to 15 radially-disposed tall, wedge-shaped columnar cells. The nuclei are regularly arranged in a small circle in the center of the cord. In longitudinal section, a cord appears to consist of two rows of tall columnar cells with a double row of nuclei running lengthwise in the center of the cord. A common occurrence in the garter snake adrenal is the presence of strands and large masses of undifferentiated cortical cells in the capsule (Fig. 25). These masses of cells, if followed in serial sections, are often found to be continuous with the parenchymal cortical tissue. The undifferentiated cells have a very limited amount of cytoplasm which, after Masson's staining, appears strongly fuchsinophilic.

The garter snake adrenal is an extremely vascular gland and this is made apparent in histological preparations by the large and extensive network of vascular sinuses that pervades the gland (Fig. 25). These sinuses are lined with a thin endothelium having extremely flattened nuclei.

Cytology

Two tinctorial types of medullary cells are found in the garter snake adrenal. The cells aggregated near the dorsally-located artery have a yellow, finely-granular appearance after Zenker-formal fixation and Masson's stain. The small clumps and individual cells associated with the cortical tissue possess coarse granules and stain reddish-brown. Both types are generally polyhedral in shape and measure 12 to 14 microns in diameter.

The cortical cells are usually of a tall columnar type measuring 20 to 25 microns in height. There is neither notable sexual variation nor discernable seasonal difference in cell height in the series of garter snakes which was collected in April, July and October. The cells exhibit a marked polarity with the nucleus usually located near the end of the cell distal to the blood supply (Figs. 25 & 28). The nuclei are 4 to 5 microns in diameter, spherical-to-oval in shape, and exhibit from one to three prominent nucleoli.

The mitochondria are granular bodies occurring throughout the cytoplasm (Fig. 28). In those cells with a high lipid concentration, the mitochondria are few in number and widely scattered in the cytoplasm surrounding the lipid vacuoles. In the lipid-poor undifferentiated cells, a heavy concentration of mitochondria pervades the cytoplasm. There is an inverse ratio between the amount of mitochondria and lipid in the cortical cells. Fuchsinophilic spheres larger than mitochondria occur frequently in the cytoplasm.

An outstanding feature of the garter snake adrenal is the distribution of lipid inclusions in the cortical cells. The lipid droplets are osmophilic, sudanophilic, Schultz-positive, and exhibit an intense positive reaction to the Schiff leucofuchsin reagent. The lipid droplets appear to be evenly distributed throughout the cortical tissue and no tendency toward zonation is observed (Fig. 26).

Within the cells, the lipid droplets are concentrated at the peripheral end of the cell and at the end opposite the nucleus. This polarity is strikingly demonstrated in frozen sections stained with Sudan black B (Fig. 26), or by the Shultz technique for demonstrating cholesterol and its esters (Fig. 27). A similar distribution of Schiff-positive carbonyl lipids is seen, there being a parallel pattern of distribution of the sudanophilic, Schultz-positive and Schiff-positive lipids. By chance, one of the frozen sections tested for cholesterol was observed to have a mass of undifferentiated cells in the capsule (Fig. 27). Only a slightly positive reaction was seen in this nodule.

(b) *Natrix sipedon*

The adrenals of the common water snake, *Natrix sipedon*, are in general similar in morphology to those of *Thamnophis*. Medullary tissue is aggregated in the dorsal region of the capsule and small groups of cells are scattered throughout the cortical tissue. The cortical tissue exhibits a regular arrangement of polarized columnar cells into anastomosing cords. The polarization is similar to that of the garter snake.

(c) *Lampropeltis doliata*

The morphology of the adrenals of the Eastern milk snake, *Lampropeltis doliata*, is similar to that of *Thamnophis* and *Natrix*.

(d) *Ophedrys vernalis*

The adrenals of the smooth green snake, *Ophedrys vernalis*, are similar in anatomical features to those of the other colubrid snakes mentioned. Histologically, however, the parenchyma of the gland is more compact. The cortical tissue is arranged in radial cords of low

columnar cells. The nuclei are located distal to the capillaries.

2. Family Crotalidae

Only two specimens of the prairie rattlesnake, *Crotalus viridis*, were studied. Both of these animals had been preserved in 10% formalin for an unknown period of time. The adrenals were poorly fixed, yet on staining with Mayer's acid hemalum and eosin, sufficient detail was present to warrant comment. Anatomically, the glands are similar to those described for the garter snake. Microscopically, a typical ophidian disposition of cell types is seen. The medullary tissue is confined primarily to the dorsal capsular region while the cortical cells are regularly arranged in anastomosing cords. The nuclei show a similar disposition and polarity to that noted for the garter snake.

3. Family Boidae

(a) *Boa constrictor*

The adrenals of the boa constrictor are similar, for the most part, to those of the colubrid snakes. There is a tendency toward an elongation of the right gonad and a consequent elongation of the right adrenal. The microscopic anatomy is the typical ophidian type with a dorsal cap of medullary cells plus small clumps and single cells scattered in the cortical tissue. The cortical tissue consists of anastomosing cords made up of radially arranged low columnar cells exhibiting a slight polarity. The nuclei are arranged near the center of the cords or distal to the blood sinuses.

(b) *Charina bottae*

The adrenals of the rubber boa, *Charina bottae*, are elongated thread-like glands located in the mesenteries supporting the gonads. The left adrenal measures approximately 10 millimeters in length and 1 millimeter in width, whereas the right adrenal is approximately 25 millimeters in length and 1 millimeter in width.

The vascular supply of these glands consists of a single artery to the left adrenal and three arteries to the right adrenal. Blood leaves the glands by a series of short efferent adrenal veins, 6 for the left adrenal and 12 for the right adrenal. The glands are not attached directly to the vena cava, as noted in other snakes, but both glands empty into the vena cava, the left adrenal lying just anterior to the junction of the efferent renal veins.

In order to determine the arrangement of the cortical and medullary tissue, histological preparations were made of glands removed from museum specimens. Formalin-fixed tissues such as these will, on staining with Weigert's iron

hematoxylin, give fairly good differentiation of cortical and medullary cells. The medullary cells exhibit a strong basophilia and stain dark blue. The adrenal has a regular ophidian distribution of glandular tissue with a dorsal aggregation of medullary cells and the cortical cells arranged in cords. In sections the cords appear to be made up of several layers or tiers of cells.

DISCUSSION

Anatomy

The adrenal glands of reptiles exhibit a consistent association with the reproductive organs and the venous system. These anatomical relationships are found in all four orders of the reptiles studied.

In the Crocodylia, the adrenals are discrete, elongate bodies located in a retroperitoneal position on the midventral surface of the kidney in close association with the ventrally situated gonads and gonaducts. This relationship has been described for the alligator (Pettit, 1896; Spanner, 1929; Reese, 1931; Lawton, 1937), and the caiman (Pettit, 1896), and is confirmed in the present study.

A consistent anatomical relationship is found in all the species of North American turtles examined in this study. The adrenals are located on the ventral surface of the kidneys in close association with the gonaducts and gonads. The latter, however, are separate from the adrenal, being suspended ventrally from the adrenal-kidney complex by the mesorchia or mesovaria. The adrenal is not as well organized into a discrete gland as is that of the Crocodylia, but, rather, usually consists of partially fused spherical-to-irregular-shaped masses intimately associated with the vascular plexus on the ventral surface of the kidney. This close relationship with the kidney resembles the condition found in the amphibians (Radu, 1931; Villee, 1943).

There are few anatomical descriptions of the adrenals of the Chelonia in the literature. Vincent (1896), Minervini (1906) and Thompson (1932) give brief descriptions of the genus *Testudo*. Ogushi (1911) describes their relationship to the kidneys in the soft-shelled turtle, *Trionyx japonicus*. Kuntz (1912) describes the embryonic development and gross anatomy of the adrenals in young specimens of the loggerhead turtle, *Caretta caretta*. The gross anatomical relationships in *Emys europaea* have been reported by Spanner (1929). These early descriptions have been reviewed by van der Spenkel (1934). In the marine tortoise, *Caretta caretta*, the adrenals are usually united into a single structure, according to Holmberg & Soler (1942).

The adrenal glands in the Sauria are found

enclosed in the mesorchia or mesovaria supporting the gonads. This consistent relationship has been reported in the agamid lizard, *Uromastix hardwickii* (Vincent, 1896), the *Chamaeleon* sp. (Minervini, 1904), various iguanids (Brooks, 1906; Reynolds, 1938), in the Laceratidae (Braun, 1882; Pettit, 1896; Vincent, 1896; Krause, 1922; Radu, 1934), the teiid lizard, *Cnemidophorus gularis* (Brooks, 1906), the anguimorph lizards, *Gerrhonotus* (Retzlaff, 1949), *Heloderma* (Hartman & Brownell, 1949) and members of the Varanidae (Pettit, 1896).

The adrenals of the Serpentes bear the same relationship to the reproductive organs as was noted in the lizards; however, in this group the elongation of the body has resulted in an asymmetrical placement of the paired internal organs. The right adrenal is always situated cephalad to the left. This arrangement has been observed in many colubrid snakes: *Natrix natrix* (Pettit, 1896; Spanner, 1929; Radu, 1934), *Coluber* sp. (Vincent, 1896; Minervini, 1904; Uchoa Junqueira, 1944), *Dryophylox* sp. (Valle & Souza, 1942), the garter snake, *Thamnophis sirtalis* (Atwood, 1918) and the water snake, *Natrix sipedon* (Bragdon, 1953).

The relative position of the adrenals in *Thamnophis sirtalis* can best be correlated with the ventral scales, in which case a consistent relationship is noted. The left adrenal lies between the 35th to 37th gastrostege in males and between the 31st to 34th in females. In both sexes, the right adrenal lies between the 45th and 48th scute, counting anteriorly from the anal plate. This separation of approximately ten scutes has been reported also for the common water snake, *Natrix sipedon* (Bragdon, 1953).

While extensive data on adrenal weights were not obtained, the average determined—0.045 per cent. of the body weight—agrees with the range of 0.04 to 0.05 per cent. reported for other colubrid snakes by Naccarati (1922).

Bourne (1936, 1949) states that the adrenals of the boa constrictor exhibit a primitive anatomical condition in that they are located along the lobes of the attenuated kidneys. This association with the kidneys, however, was not found in the specimens of the boa constrictor and the rubber boa, *Charina bottae*, that were examined in this study. Both species have a typical serpentine arrangement with the adrenals located along the greatly elongated and asymmetrically-placed gonads. This disposition of the adrenals has previously been reported for the boa constrictor by Poll (1905) and the python (Pettit, 1896; Beddard, 1906a, 1906b; Spanner, 1929). In the very young anaconda, *Eunectes notaeus*, Beddard (1906a) noted that the adrenals are associated with the persistent mesonephros but

that this condition is not found in adult animals. This would suggest that Bourne probably based his descriptions on very young animals that still possessed mesonephros kidney tissue.

The vascular supply of the adrenals of reptiles consists of both an arterial and venous supply. In the Crocodylia, the arterial supply consists of several pairs of arteries which arise from the dorsal aorta and enter the dorso-mesial region of the capsule. This general picture has been described by Pettit (1896), Reese (1931) and Lawton (1937). The arterial supply of the adrenal in the Chelonia consists of several small paired adrenal arteries from the aorta, as well as small branches from the renal and genital arteries. Very few observations on the arterial supply have been reported in the literature. Thompson (1932) described the arterial supply to the adrenal in the tortoise, *Testudo graeca*.

The arterial supply to the adrenals in the Sauria exhibits some variation among the various species studied. In the horned lizard, *Phrynosoma cornutum*, each adrenal receives one (sometimes two) arteries from the dorsal aorta. These arteries also supply the gonads and perhaps should be referred to as the spermatic or ovarian arteries. The adrenals receive blood from branches arising from these arteries, as the former are in close contact with the adrenals. In *Anolis carolinensis*, separate arteries supplying the adrenal are present. Pettit (1896) reported finding three arteries supplying the adrenal in *Varanus niloticus* and only a single artery to each gland in *Varanus salvator*. Bhatia (1929) observed that branches of the spermatic arteries supply the adrenals of the agamid lizard, *Uromastix hardwickii*. According to Retzlaff (1949), the arterial adrenal blood in the alligator lizard, *Gerrhonotus multicarinatus*, is derived from branches from the aorta, renal and mesenteric arteries.

In the Serpentes, the adrenal arterial supply comes from branches of the spermatic or ovarian artery as it courses in an anterior-posterior direction along the dorsal region of the connective tissue capsule. This general pattern has been reported for *Natrix natrix* (O'Donoghue, 1912), for the rat snake, *Ptyas mucosus* (Ray, 1934), and previously reported for the garter snake, *Thamnophis sirtalis*, by Atwood (1918).

A unique feature of the vascular supply of the reptilian adrenal is the presence of an adrenal portal system. This system of vessels has been carefully studied by a number of morphologists and their observations have been reviewed by van der Sprenkel (1934). In the alligator and caiman, a single adrenal portal vein arises from the dorsal body wall anterior to the adrenal,

enters the anterior pole of the adrenal, and branches into a plexus connecting with the vascular sinuses in the parenchyma of the gland. This arrangement has been described by Pettit (1896), Lawton (1937) and Spanner (1929) and is confirmed in the histological preparations of this study.

In the Chelonia, Spanner (1929) pictures three adrenal portal veins branching off from the renal portal veins. The presence of a portal system in the turtles which were studied could not be definitely determined by the histological approach employed.

In the Sauria, a variable number of portal veins branch off from the dorsal body wall and enter the adrenals. Beddard (1904b, 1904c, 1905) and Spanner (1929) examined a number of species of lizards and noted considerable species variation in the number and relationship of the adrenal portal veins. In the present study, a single adrenal portal vein was observed entering the anterior pole of the adrenal of *Phrynosoma cornutum*. Retzlaff (1949) does not mention a portal supply for *Gerrhonotus* nor does Miller (1952) for *Xantusia*.

In the Serpentes, the adrenal portal system is well developed. For example, in the garter snake, *Thamnophis sirtalis*, two relatively large veins enter each adrenal from the dorsal body wall. From one to three veins have been reported entering the snake adrenal (O'Donoghue, 1912; Ray, 1936; Beddard, 1904a, 1904d, 1906a).

According to Beddard (1906a), the adrenal portal veins in reptiles are persistent parietal branches of the postcardinal veins associated with the embryonic mesonephros kidneys. Their adrenal portal relationship is a secondary function taken over in the mature animal.

It is interesting to note that the adrenals of reptiles are always located in the region where the efferent renal veins join to form the inferior vena cava. In the Chelonia, both adrenals are in intimate contact with the efferent renal veins. In the Crocodylia, the adrenals are associated with the vena cava, the right adrenal being more intimately joined to the vena cava than the left. In the Sauria and Serpentes, the left adrenal usually drains either via short efferent veins or directly into the anterior portion of the left efferent vein while the right adrenal usually drains directly into the vena cava.

The nerve supply to the adrenal is derived from lateral sympathetic ganglia, the extent of innervation differing somewhat among the various orders of reptiles. In the Crocodylia, Lawton (1937) described the innervation of the alligator adrenal as arising from four to five ganglionic clusters. Fibers from these ganglia enter the gland and associate themselves with

medullary tissue. This segmental pattern of innervation was found in both the alligators and caiman examined during this study.

The innervation of the chelonian adrenal is more complex than that of the other reptiles. Because of the close association of the gland with the kidney, it is difficult to determine the exact relationship of the numerous ganglia which are located in or near the adrenal.

In the lizards and snakes, sympathetic ganglia are found chiefly on the dorsal surface of the gland in association with the arterial blood vessels.

The exact nature of the numerous bundles of nerve fibers seen coursing in the capsule and between the parenchymal units is extremely difficult to ascertain from histological preparations. Sudan-stained frozen sections reveal thin sudanophilic sheaths in some of the nerve bundles, which is indicative of the presence of both myelinated preganglionic and unmyelinated postganglionic fibers in the adrenals of reptiles.

The medullary cells receive an abundant supply of unmyelinated fibers which end as dense networks around the cells. Kolossow (1930) observed that the nerve fibers terminate as oval patches on the medullary cells of the turtle, *Emys europaea*. In the alligator lizard, *Gerrhonotus multicarinatus*, Retzlaff (1949) noted that fine nerve fibers terminate on the medullary cells. In the alligator, painted turtle and garter snake, nerve fibers are found running along between the cortical strands and often fine side-branches are given off at right angles, which appear to penetrate into the cortical tissue. The innervation of the cortical cells is a moot question. Some investigators claim that nerve fibers terminate on the cortical cells (Alpert, 1931, in the human; and Retzlaff, 1949, in the alligator lizard). On the other hand, Hollinshead (1936) and Swinyard (1937) found no evidence of cortical tissue innervation in the cat adrenal. Also, Kolossow (1930) was not able to discover nerve endings to cortical cells of the turtle, *Emys europaea*.

The identification of argyrophilic nerve fibers with specific cortical cells is a difficult problem inasmuch as the fibers may not be destined for the cortical cells, but rather for some other nearby tissue such as blood vessels. Swinyard (1937) cautions that the silver techniques employed may possibly stain reticular fibers which may be incorrectly identified as nerve fibers.

Histology

The reptilian adrenal gland is enclosed in a thin connective tissue capsule continuous with the enveloping mesentery or surrounding connective tissue. The capsule is, in general, com-

posed of an outer layer of collagenous fibers continuous with the adjacent connective tissue and an inner layer of reticular fibers continuous with the reticular framework supporting the glandular parenchyma. Elastic fibers are confined to the media of blood vessels that are found in the capsule and trabeculae. The relative amount of collagenous and reticular fibers in the gland varies with both the species and the age of the animal. In young animals, the parenchyma is supported by a thin network of reticular fibers while in old adult animals the reticular framework is supplemented by thick collagenous fibers, the amount of which varies from species to species. In adult turtles, there is considerable infiltration of thick collagenous trabeculae from the capsule, which tend to partition the cortical tissue.

Some species variation is found in the lizards. For example, Retzlaff (1949) observed in *Gerrhonotus* heavy bundles of collagenous fibers surrounding the cortical cells, with fine fibrils originating from these bundles passing among the cells. This pattern is also found in the anguimorphid lizard, *Ophisaurus*, examined in this study, but it is not present in the Scincidae. In the skink, *Eumeces fasciatus*, the cortical cords are enclosed in a network of collagenous fibers; however, fibers do not appear to penetrate among the individual cells comprising a cord.

An interesting and significant departure from the hereinbefore described parenchymal stroma is found in the garter snake. The cortical cords are usually enclosed in a loose connective tissue network composed chiefly of collagenous fibers. This collagenous network is infiltrated with numerous vascular sinuses supported by a fine reticular network. This same arrangement has been reported for the European snake, *Coluber* sp., by Minervini (1904).

The reptilian adrenal gland consists of three kinds of cells—cortical, medullary, and sympathetic nerve cells—arranged in definite patterns which appear to be characteristic for the major phylogenetic groups. In the Crocodylia, the medullary tissue consists of an encircling peripheral zone in addition to small clumps, strands and individual cells scattered in the interspaces and sinuses between the columns of cortical cells. This general crocodylian pattern is found in the alligator and caiman. Reese (1931) and Lawton (1937) have also described this arrangement in the alligator; however, no descriptions of Old World crocodiles were found in the available literature.

A random intermixing of the medullary and cortical components is typical of the Chelonia. The medullary tissue is not concentrated in any particular region of the gland but rather is ir-

regularly distributed among the cortical cell groups as clusters and strands of medullary cells. These clusters are of irregular size and shape, tending to exhibit some species differences. For example, the spotted turtle, *Clemmys guttata*, is unique in having larger and more numerous clumps of medullary cells than those observed in other species of turtles studied. This general chelonian arrangement has been described for several species of turtles: *Testudo tabulata* and *Malacllemys terrapin*, (Vincent, 1896), and *Emys europaea* (Poll, 1906; Radu, 1934). Moreover, a similar random disposition of medullary and cortical components is also present in the avian adrenal (Hartman, et al., 1947).

In the Squamata (lizards and snakes), the medullary tissue is aggravated principally on the dorsal side of the cortical tissue with the degree of intermingling with the cortical tissue varying from species to species. In the horned lizard, *Phrynosoma cornutum*, there is considerable infiltration, while in *Anolis carolinensis* little medullary tissue is found in the cortical parenchyma. This scarcity is also true of the skinks, *Eumeces obsoletus*, *E. fasciatus* and *Lygosoma laterale*. Among the anguimorphid lizards, Retzlaff (1949) reported that the medullary cells of the alligator lizard, *Gerrhonotus multicarinatus*, are intermingled with the cortical portion. This was also noted in this study for *Gerrhonotus coeruleus* as well as for the related species, *Ophisaurus ventralis*. In addition, Hartman & Brownell (1949) noted some intermingling in the Gila monster, *Heloderma suspectum*.

In the Serpentes, the medullary cells are chiefly aggregated in a compact unit associated closely with arterial blood vessels located on the dorsal side of the capsule. A more or less uniform scattering of small medullary clusters of one to four cells is found in the cortical parenchyma. This arrangement is observed in the garter snake, water snake and other colubrid snakes examined, as well as in the boa constrictor.

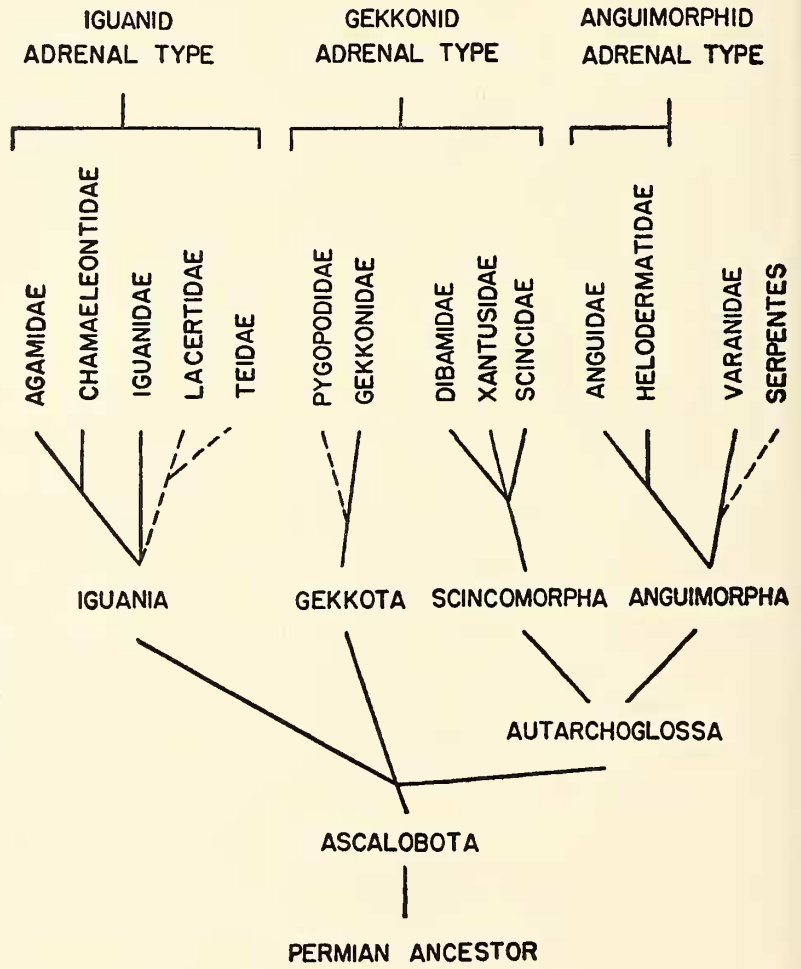
The cortical tissue exhibits considerable variation in shape and arrangement of the cells, with an appearance of a more or less characteristic pattern for each species. In the Crocodylia, namely, the alligator and caiman, the cortical tissue consists of anastomosing cords composed of polyhedral-to-tall columnar cells having a more or less regular arrangement. Probably because of compression, the outer cords in the adrenal often appear to have a more regular cellular arrangement. In this instance, the peripheral cords may consist of a single row of greatly elongated and flattened cells, as noted for the alligator, or as a double row of cells, in the case of the caiman.

In the Chelonia, the cords usually consist of

irregularly-shaped cells which, in longitudinal section, consist of several rows of cells having very indistinct cell outlines and an abundant and irregular distribution of nuclei. The more centrally located columns usually consist of more than two tiers of cells; on the other hand, peripheral cords (as a result of compression) often consist of a single row of greatly elongated and flattened cells. In the family Kinosternidae, the cells tend to have a more regular arrangement, with the cells appearing to be radially disposed in the cords.

The disposition of the cortical cells in the Sauria is of particular interest for the reason that certain phylogenetic trends are apparent. On arranging the various species of lizards (described in this study and elsewhere in the literature) into groups on the basis of the disposition and morphology of the cortical cells, they fall into three general categories having similar histological features: the gekkonid, anguimorphid and iguanid types.

These histological trends appear to substantiate the suggested phylogenetic relationships of the various lizard groups. According to Camp (1923), the Sauria constitute a natural and diversified group arising from some unknown Permian stem reptile which soon divided into two main groups, the Ascalobota and the Autarchoglossa. The former includes the Gekkonidae, Agamidae, Chamaeleontidae and Iguanidae, with the Gekkonidae considered to be the most primitive family. The Autarchoglossa are subdivided into the Scincomorpha, which includes the Xantusidae, Scincidae, Lacertidae and Teidae, and the Anguimorpha, which includes the Anguidae and the platynotid lizards, of which the Varanidae are a part. The Xantusidae are believed to be an intermediate group between the Ascalobota and Autarchoglossa. These suggested relationships are shown in Text-fig. 1, modified after Camp (1923). Only those families are included for which histological data are available for comparison.



TEXT-FIG. 1. Comparison of adrenal histological types with suggested phylogenetic relationships of the lizards. (Phylogenetic chart modified from Camp, 1923).

The characteristic histological feature of the gekkonid type of adrenal is the radial arrangement of tall wedge-shaped columnar cells into cords having a consistent disposition of the nuclei at the periphery of the cords. This arrangement is found in the four species of the Gekkonidae described in this study as well as in the three species belonging to the family Scincidae. The gekkonid pattern is also observed in the Dibamidae and Pygopodidae, and has been described in the Xantusidae by Miller (1952).

The anguimorphid pattern consists of an irregular arrangement of polyhedral low columnar cells with no general tendency to form cords consisting of regularly arranged tiers of cells. This arrangement is found in the glass snake, *Ophisaurus ventralis*, and the closely related species, *Gerrhonotus coeruleus*. Retzlaff (1949) has also described this irregular disposition of cortical cells in the latter genus. Hartman & Brownell (1949) found a similar pattern to exist in another anguimorphid lizard, the Gila monster, *Heloderma suspectum*.

The disposition and morphology of the adrenal cortical cells in the Iguania is somewhat variable, even within family groups; however, all the species that have been described appear to have a more or less regular arrangement of the cells into cords. In the Agamidae, Vincent (1896) describes the cortical tissue of *Uromastix hardwickii* as comprised of cords of cells consisting of a double row of tall columnar cells each having a centrally placed nucleus. Minervini (1904) pictures the cortical cords of the *Chameleon* sp. as composed of small polyhedral cells with centrally located nuclei. In the Iguanidae, the cortical cells of *Anolis carolinensis* and *Sceloporus undulatus* would fit more or less the above description. On the other hand, cortical cells of the horned lizard, *Phrynosoma cornutum*, are more inclined to be of a tall columnar type with the nuclei tending toward a peripheral position.

While certain anatomical similarities suggest that the Lacertidae and Teiidae are closely related to the Scincidae (Camp, 1923), the histological features of the adrenal gland of these two families resemble more closely the iguanid type rather than the gekkonid pattern.

According to Camp (1923), the Gekkonidae represent the most primitive group with the Xantusidae being intermediate between the Gekkonidae and the Scincidae. The Lacertidae are considered an offshoot from the Scincidae and the Teiidae as a branch of the Lacertidae. The Anguimorpha are believed to have separated from the Scincomorpha very early in the Mesozoic era.

If, as Camp points out, the Gekkonidae re-

present the most primitive group, then it would seem justifiable to consider that the gekkonid type of adrenal found in the Gekkonidae represents a primitive morphological condition. This type is also present in the following groups: Xantusidae, Scincidae, Dibamidae and Pygopodidae. The Iguania, on the other hand, have diverged from the gekkonid pattern, showing only an occasional similarity as, for example, in *Phrynosoma cornutum*. Furthermore, the Anguimorpha (Anguidae and Helodermatidae) show little similarity to the gekkonid or iguanid types and have in their long geologic separation either undergone an evolutionary modification from the primitive gekkonid pattern or else may have been derived from primitive Ascalobota differing entirely from the primitive Gekkonidae.

The cortical cells of the Serpentes are arranged into cords having a characteristic pattern. In the garter snake, *Thamnophis sirtalis*, the cells are radially arranged into cylindrical cords which exhibit a striking regularity and polarity. In longitudinal sections of a cord, the cells are disposed in two rows of tall columnar cells having a double row of nuclei aligned lengthwise near the center of the cord. In transverse section, a cord consists of a radially arranged group of tall, wedge-shaped cells having a centrally located ring of nuclei. This consistent pattern is also found in the water snake, *Natrix sipedon*, and the rattlesnake, *Crotalus viridis*. In the rubber boa, *Charina bottae*, the cords appear to be made up of several tiers of cells having a more irregular arrangement.

Cytology

(a) Medullary cells

Two types of medullary cells can be distinguished in all of the species of reptiles studied on the basis of their tinctorial characteristics: a "light" type of cell usually light yellow in color and having fine granules; and a yellowish-brown "dark" type having coarse granules. The degree of staining intensity and the relative number of each type varies from order to order and from species to species. The two cell types are best distinguished after fixation in solutions containing chromium salts, such as Zenker's, followed by stains containing acid fuchsin (for example, Masson's). Classical literature often refers to the "chromaffin reaction" as a selective staining of the cytoplasmic granules by the chrome salts; however, Coupland (1953) points out that the yellow-to-brown color is the result of the oxidation of a certain amount of catechol amines into corresponding quinones and other complex oxidation products, such as pigments.

The occurrence of different medullary cell types has been reported in mammals (Bänder,

1951; Bourne, 1949); birds (Müller, 1929); reptiles (Radu, 1934; Reese, 1931; Retzlaff, 1949; Miller, 1952); and amphibians (Fustinoni & Porto, 1938).

(b) Cortical cells

The most characteristic feature of the cortical cells is the presence of lipid droplets and granular mitochondria, there being an inverse relationship between the amount of each of these cytoplasmic inclusions present. Histochemical studies indicate that the droplets are a mixture of neutral fats, cholesterol and cholesterol esters, and carbonyl-containing lipids. This description portrays, in general, the fundamental functional unit of the cortical tissue found throughout the various vertebrate classes: shark (Dittus, 1941; Hayes, 1941), lungfish (Holmes, 1950; Gérard, 1951), frog (Radu, 1931), bird (Müller, 1929; Miller & Riddle, 1942; Knouff & Hartman, 1951) and mammal (Greep & Deane, 1949; Nicander, 1952).

The number of lipid droplets may vary slightly from cell to cell; however, there is no tendency toward a zonation similar to that found in the mammals. On the other hand, the distribution of the lipid droplets within the cells varies significantly among the different species of reptiles that were studied, and it is possible to distinguish at least three basically different cytological cell types.

The cortical cells of the garter snake are of special interest for they exhibit a polarization in which the lipid droplets are localized at the free end of the cell in closest contact with the blood stream, while the nucleus is placed near the base of the cell. The granular mitochondria are scattered in the interstices between the lipid droplets. This unique disposition of the lipid droplets, mitochondria and nucleus suggests that an exchange of materials occurs at the peripheral surface adjacent to the blood supply while presecretory synthetic processes occur in the region between the nucleus and the free end of the cell. A similar type of functional polarity has been described for the brown pelican (Knouff & Hartman, 1951).

A functional polarity the reverse of that just mentioned is found in certain species of lizards. In the skink, *Eumeces obsoletus*, the lipid droplets and mitochondria are concentrated near the base of the cells with the nucleus located at the peripheral edge next to the blood stream. This interesting cytological arrangement is also found in other species of skinks, geckos, xantusids, dibamids and pygopodids. A satisfactory explanation for this unique functional adaptation cannot be derived from histological studies alone. Miller (1952) found that the lipid content of the

cortical cells of *Xantusia* can be experimentally altered; however, the mechanism as to how secretory products reach the blood stream still remains unknown.

The third functional type has the nucleus centrally located and exhibits an even distribution of lipid droplets and mitochondria throughout the cytoplasm. This disposition is found in the cortical cells of the alligator, painted turtle and green anolis.

SUMMARY

1. The adrenal glands of reptiles exhibit a consistent association with the reproductive organs.

2. The vascular supply consists of both arterial and venous blood with the origin and number of vessels supplying the adrenals varying from species to species. A close association with the efferent renal veins and vena cava is common to all species.

3. The nerve supply to the adrenal is derived from lateral sympathetic ganglia with the extent of innervation differing somewhat among the various orders of reptiles. The medullary component of the gland is innervated by networks of unmyelinated fibers; however, only a few fibers are found in the cortical tissue.

4. The reptilian adrenal gland is enclosed in a thin connective tissue capsule composed of an outer layer of collagenous fibers continuous with the adjacent connective tissue and mesenteries, and an inner layer of reticular fibers continuous with the reticular framework supporting the parenchyma of the gland. The relative amount of collagenous and reticular fibers in the gland varies with both the species and the age of the animal.

5. The reptilian adrenal gland consists of three kinds of cells—cortical, medullary and sympathetic nerve cells—arranged in definite patterns which appear to be characteristic of the major phylogenetic groups.

6. In the Crocodylia, the medullary tissue consists of an encircling peripheral zone in addition to small clumps, strands and individual cells scattered in the cortical parenchyma. A random intermixing of the medullary and cortical components is typical of the Chelonia. In the Sauria and Serpentes, the medullary tissue is aggregated principally on the dorsal side of the cortical tissue, with only a slight intermixing.

7. The cortical tissue exhibits considerable variation in shape and arrangement of the cells with the appearance of a more or less characteristic pattern for each species.

8. The disposition of the cortical cells in the Sauria is of particular interest for the reason that certain phylogenetic trends are apparent.

The Sauria can be divided into three general groups: the gekkonid, anguimorphid and iguanid types, on the basis of similar histological features.

9. Two types of medullary cells can be distinguished in all of the species of reptiles studied on the basis of their tinctorial characteristics.

10. The cortical cells are characterized by the presence of lipid droplets and granular mitochondria, there being an inverse ratio between the amount of each of these present. Histochemical studies indicate that the droplets are a mixture of neutral fats, cholesterol esters and carbonyl-containing lipids. The number of lipid droplets varies from cell to cell; however, there is no tendency toward a zonation.

11. Polarity differences in the distribution of the lipid droplets, mitochondria and nuclei in the cortical cells of the various species studied illustrate fundamental variations in the cytology of cortical cells.

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EXPLANATION OF THE PLATES

PLATE I

- FIG. 1. Transverse section through alligator adrenal (α) showing relationship to kidneys (k), gonads (g), dorsal aorta (d), vena cava (v) and sympathetic ganglia (s). Bouin's fixation and Masson's stain. $\times 24$.
- FIG. 2. Transverse section through adrenal of alligator showing sympathetic nerve fibers in capsule, peripheral zone of "light" medullary cells, strands of "dark" medullary cells extending into cortical tissue and irregular shape and arrangement of light-staining cortical cells. Zenker-formal fixation and Masson's stain. $\times 130$.
- FIG. 3. Transverse section through adrenal of alligator showing distribution of reticular network supporting parenchyma of gland. Note heavy reticular fibers running lengthwise with cords and fine network of reticular fibers that encircles the cortical cords. Bouin's fixation and Foot's modification of Hortege's silver carbonate technique. $\times 240$.
- FIG. 4. Frozen section of alligator adrenal showing distribution of sudanophilic lipids in the cortical cells. Note absence of stain in medullary cells. Baker's formalin fixation and Sudan black B stain. $\times 240$.
- FIG. 5. Transverse section of alligator adrenal showing centrally located nuclei and the uniform distribution of granular mitochondria in the lipid-rich cortical cells.

Note cluster of "dark" medullary cells at upper left and strand of "light" medullary cells at right-hand edge. Zenker-formal fixation, post-chromated for 48 hours and stained with aniline-acid fuchsin and methyl blue. $\times 690$.

PLATE II

- FIG. 6. Transverse section through adrenal (α) of painted turtle, *Chrysemys picta*, showing relationship to kidneys (k), gonads (g), dorsal aorta (d) and vena cava (v). Zenker-formal fixation and Masson's stain. $\times 24$.
- FIG. 7. Transverse section through adrenal of painted turtle showing disposition of medullary and cortical tissue. Note irregular arrangement of cortical cells into anastomosing cords. Zenker-formal fixation and Mayer's acid hemalum and eosin. $\times 230$.
- FIG. 8. Transverse section through adrenal of painted turtle showing close association of medullary cells and sympathetic ganglion cells. Zenker-formal fixation and Mayer's acid hemalum and eosin. $\times 230$.
- FIG. 9. Frozen section of adrenal of painted turtle showing distribution of sudanophilic lipids in cortical tissue. Note single sudanophilic nerve fiber in sympathetic nerve at upper edge of photograph. Baker's formalin fixation and Sudan black B stain. $\times 240$.
- FIG. 10. Transverse section of adrenal of painted turtle showing distribution of granular mitochondria in lipid-rich cortical cells. Note centrally located nuclei in cortical cells and clump of "dark" medullary cells at upper edge of photograph. Zenker-formal fixation, post-chromated for 48 hours and stained with aniline-acid fuchsin and methyl blue. $\times 690$.

PLATE III

- FIG. 11. Oblique section through adrenal (α) of horned lizard, *Phrynosoma cornutum*, showing association with vas deferens ($v.d.$). Note peripheral zone of darker-staining medullary tissue as well as scattered masses among the cortical cells. Zenker-formal fixation and Masson's stain. $\times 24$.
- FIG. 12. Transverse section of adrenal of horned lizard showing sympathetic ganglion cells near artery. Note peripheral zone of "light" medullary cells and clumps of "dark" medullary cells lying between the cortical tissue cords. Note regular arrangement of tall columnar cortical cells. Zenker-formal fixation and Masson's stain. $\times 230$.
- FIG. 13. Frozen section of adrenal of horned lizard showing intense sudanophilia of cortical cells. Note negative-staining sympathetic ganglion. Baker's formalin fixation and Sudan black B stain. $\times 240$.

FIG. 14. Transverse section of adrenal of lizard, *Anolis carolinensis*, showing dorsal sympathetic ganglion and medullary tissue. Note close association of cortical tissue with vena cava and cord-like arrangement of cortical cells. Zenker-formal fixation and Mayer's acid hemalum and eosin. $\times 230$.

FIG. 15. Transverse section through adrenal of alligator lizard, *Gerrhonotus coeruleus*, showing pigmented capsule, peripheral zone of medullary cells, peripheral zone of "light-staining" cortical cells and irregular arrangement of cortical cells in the parenchyma of the gland. Bouin's fixation and Masson's stain. $\times 230$.

FIG. 16. Transverse section through adrenal of the glass snake, *Ophisaurus ventralis*, showing aggregation of medullary cells, large sinus opening into ventral vein and the irregular arrangement of the cortical cells. Bouin's fixation and Mayer's acid hemalum and eosin. $\times 230$.

FIG. 17. Transverse section through adrenal of the lizard, *Cnemidophorus tessellatus*, showing dorsal aggregation of medullary cells and irregularly arranged cortical cells. Bouin's fixation and Mayer's acid hemalum and eosin. $\times 230$.

PLATE IV

FIG. 18. Transverse section through adrenal of the skink, *Eumeces obsoletus*, showing typical radial arrangement of the tall columnar cortical cells into anastomosing cords. Note disposition of nuclei at the peripheral edge of the cords. Zenker-formal fixation and Masson's stain. $\times 230$.

FIG. 19. Transverse section of adrenal of skink, *Eumeces fasciatus*, showing peripheral position of nuclei and scarcity of granular mitochondria in lipid-rich cytoplasm of cortical cells. Zenker-formal fixation, post-chromated for 48 hours and stained with aniline-acid fuchsin and methyl blue. $\times 690$.

FIG. 20. Frozen section of adrenal of skink, *Eumeces obsoletus*, showing characteristic localization of sudanophilic lipids in central region of cortical cords and clear peripheral nuclear zone. Baker's formalin fixation and Sudan black B stain. $\times 240$.

FIG. 21. Transverse section through the adrenal of the gecko, *Hemidactylus turcicus*, showing dorsal medullary tissue and cords of radially arranged tall columnar cortical cells with peripherally located nuclei. Zenker-formal fixation and Masson's stain. $\times 230$.

FIG. 22. Transverse section through adrenal of the gecko, *Tarentola mauritanica*, showing similar disposition of cortical cells as noted for *Hemidactylus* and *Eumeces*. Zenker-formal fixation and Masson's stain. $\times 230$.

FIG. 23. Transverse section through adrenal of skink, *Lygosoma laterale*, showing dorsal medullary tissue and cords of radially arranged tall columnar cortical cells with peripherally located nuclei. Zenker-formal fixation and Masson's stain. $\times 230$.

PLATE V

FIG. 24. Transverse section through the adrenal (**a**) of the garter snake, *Thamnophis sirtalis*, showing relationship to ovary (**o**) and mesovarium (**mes.**). Note ovarian artery (**o.a.**) in dorsal capsular region and intimate association of adrenal with the ventrally located vena cava (**v**). Bouin's fixation and Masson's stain. $\times 28$.

FIG. 25. Transverse section through the adrenal of the garter snake showing large capsular nodule of undifferentiated cortical cells, dorsal aggregation of medullary cells at right of nodule and cords of radially arranged tall columnar cortical cells with nuclei arranged in regular rows at center of cord. Note small groups of dark-staining medullary cells dispersed between the

cortical cords and the extreme vascularity of the gland. Bouin's fixation and Masson's stain. $\times 120$.

FIG. 26. Frozen section of garter snake adrenal showing characteristic localization of sudanophilic lipids at periphery of cortical cords and clear nuclear zone in center of cords. Baker's formalin fixation and Sudan black B stain. $\times 240$.

FIG. 27. Frozen section of garter snake adrenal showing distribution of Shultz-positive substances. Note slight staining of undifferentiated cortical cells in capsular connective tissue. Baker's formalin fixation and Shultz cholesterol technique. $\times 240$.

FIG. 28. Transverse section of garter snake adrenal showing characteristic arrangement of tall columnar cortical cells. Note centrally located nuclear zone and uniform distribution of granular mitochondria in lipid-rich cytoplasm. Zenker-formal fixation, post chromated for 48 hours and stained with aniline-acid fuchsin and methyl blue. $\times 690$.